VII. Some Points in the Structure and Development of Dentine.

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[PLATES 36-39.]

THE difficulties attendant upon the accurate microscopical investigation of enamel and dentine, especially of the latter tissue, have been so great, that very varied views as to their structure and development have been held up to the present time.

In the study of the development of dentine, where it is necessary to retain a very soft and delicate tissue, the pulp, in its natural relations to a hard, calcified tissue, the dentine, the difficulties have been greatest, and it has generally been found necessary to resort to decalcification of the hard tissue by acids, to ensure this retention of the natural relations of the parts.

Even, however, by this method, aided by the improved processes of section cutting now in use, good preparations, in which the cells of the pulp are retained in undisturbed contact with the dentine through any great extent of surface, are comparatively rare, and the cells of the pulp usually exhibit by their shrunken condition the injurious effects of the reagents used.

It is probable also that there are other detrimental effects of these reagents present, which are not so easily detected, and the fact of the lime salts being removed from the dentine, does, as I shall endeavour to show, produce an alteration in its microscopical appearances.

By a process elaborated by Dr. L. A Weil, of Munich, and suggested to him by a method employed by Professor von Koch, of Darmstadt, in the preparation of Mollusca, the last-named difficulties have been overcome, and we can now cut preparations of the teeth with the pulps in situ, without decalcification. The process was described by Dr. Weil in a paper entitled 'Zur Histologie der Zahnpulpa' (Leipzig, 1887), and in the 'Zeitschrift für Wissenschaftliche Mikroskopie' (vol. 5, 1888, pp. 200–202), also by myself in the 'Transactions of the Odontological Society of Great Britain' (vol. 22, May, 1890).

The process consists, first, in "fixing" the soft parts by placing the freshly

extracted tooth in a saturated solution of corrosive sublimate in water—first dividing the specimen with a fine saw at one end to allow the solution to penetrate.

When sufficiently fixed, the sublimate is removed by washing, and the tooth passed gradually through successive strengths of alcohol to absolute alcohol—it is then passed into chloroform, to which are gradually added fragments of dessicated Canada balsam, until a very thick solution of the balsam is produced. The preparation is then placed in a suitable receptacle over a water-bath, covered with more of the dessicated Canada balsam, and kept at a temperature of 90° C. for several days, to allow the thorough penetration of the tissues with the hardened balsam.

The tooth is then removed, cut with a fine sharp saw under water, and the sections ground down, first on a lathe with corundum, and afterwards on a fine stone with water, under the finger. Staining in bulk may be accomplished during the treatment with alcohol, and the sections are mounted in chloroform balsam.

Very thin sections can be cut by this process, and good preparations exhibit a section of the whole pulp in undisturbed relation to the dentine, and, I think, with less alteration in the soft parts, than with any other process which has been made use of.

In the following investigation, the method above described has been employed, compared with, and controlled by, other specimens prepared by the more usual methods.

Of the various theories that have been held as to the development of dentine, the view most widely received, and embodied in all the principal text-books, is the conversion view of Tomes, Waldeyer, Boll, and others, and given by Mr. Charles Tomes, F.R.S., in his 'Dental Anatomy.'

Certain appearances in fully formed, and in developing dentine, are not thoroughly accounted for on this theory, and I propose in the following communication to describe these appearances, as demonstrated in human dentine, in other Mammalian dentines, and in the vaso-dentine of fish.

These appearances I look upon as indicative of dentine tissue being formed by a connective tissue calcification, and thus being much more closely analogous in its manner of development to bone than has usually been supposed. Other observers have described appearances not fully accounted for under the current theories of dentine development, as will appear from the following brief summary, but no observer, so far as I am aware, has either described or figured specimens in such a way as to be at all conclusive; indeed, much that has been written on the subject scarcely amounts to more than an expression of a priori hypotheses.

Of the two different theories held, as to the method by which bone is formed from cells, the "conversion' view and that of "secretion," the normer has been the most generally received.

According to the "conversion view" the masses of individual osteoblasts are consolidated by the deposit of lime salts in their substance, and the laminæ of the

bone are made up of these layers of consolidated cells—the bone corpuscles are osteoblasts, which have become included in the calcified matrix, but have themselves remained uncalcified.

The other view is that of "secretion"—the osteoblasts are held to secrete a substance which calcifies, and this secretion being not uninterrupted, but intermittent, the laminæ of bone are produced.

The layers of osteoblasts lying against the bone secrete the calcifying material, not becoming involved in the matrix themselves, with the exception of those which become enclosed by the forming bone and persist as lacunal cells.

These two different views, with some modifications, have also been held as to the formation of dentine.

JOHN HUNTER held that the dentine was formed by a process of secretion, as the shell is by the animal that forms it. In 'The Natural History of the Human Teeth,' p. 42, he says: "The ossified part of a tooth would seem to have much the same connection with the pulp as a snail has with its shell."

PURKINJE and RASHKOW held that the basis substance of the dentine originates from fibres which are formed by the dentine pulp—the dentinal canals representing the spaces between these fibres.

Schwann considered that the fibres in human teeth run in the same direction as the canals, while Purkinje and Rashkow considered the fibres to be at right angles to the tubes (parallel to the surface of the pulp), and that the dentine was deposited in successive layers.

SCHWANN says: "We have to regard the dentine as composed of fibres (basis substance) between which the canals, possessed of separate walls, are present. But," he asks, "in what relations stand the fibres and the tubes to the cells?" (odontoblasts), and he believes it possible that the dentine is the ossified substance of the pulp.

KÖLLIKER, LENT, HERTZ, BAUME, look upon the formation of dentine as a secretion process, BAUME holding that "the odontoblasts secrete a material which calcifies, rather than that they are themselves converted." WALDEYER, TOMES, BEALE, BOLL, and KLEIN, on the other hand, support the conversion view of dentine development.

Waldever, considering the process of ossification to be identical with that of ordinary bone, holds that the dentinal fibres are the central remains of the odonto-blasts, while their peripheral portions become basis substance.

Tomes ('Dental Anatomy,' p. 170) says; "The dentine is, I believe, formed by the direct conversion of the odontoblast cells, just as the enamel is by that of the enamel cells, and is derived from them and from them alone."

In a paper on the development of dentine and enamel read before the International Medical Congress, at Berlin, in August, 1890, Dr. MICHAEL MORGENSTERN regards the hardening of the tooth as a transudation process, caused by a substance in the pulp itself which contains salts of lime independent of the odontoblasts, which sub-

stance is taken up by the odontoblasts, accumulates in them, and passes out of their peripheral border, &c., &c.

Professor Klein describes the dentine previous to its calcification as showing "just like the substance of the odontoblasts the fine network of its matrix." ('Atlas of Histology.') Again, in the 'Elements of Histology,' the same authority says: "The dentine is composed of 1st, a homogeneous matrix; this is a reticular tissue of fine fibres impregnated with lime salts, and thus resembling the matrix of bone," &c., &c. Professor Klein holds that the network of reticular tissue in the substance of the odontoblasts is the reticular basis of the dentine matrix, which is thus an intracellular substance. By the aid of the mode of preparation of specimens above described, checked by the employment of other methods, I hope that I may be able to adduce more positive evidence of the inter-cellular nature of the dentine matrix than has yet been given, and to furnish something approaching to definite proof of the views herein set forth.

A transverse, or longitudinal section of the pulp and dentine of a young healthy tooth, in which dentine formation is in active progress, cut by the process described above, shows the several parts with great distinctness. (Plate 36, figs. 1 and 2.) Owing to the retention of the lime salts, the fully calcified dentine is seen very clearly differentiated from the next layer (fig. 1, b), that "tissue on the borderland of calcification" (Tomes) which is situated between the fully calcified dentine and the odontoblast layer. The layer (b) which takes the stain faintly is traversed by the dentinal fibril and encroached upon above by the advancing line of calcification (a) in the form of coalescing globules and detached spherical masses.

The odontoblast layer (c) is in immediate contact with the above-described semicalcified layer (b) and the cells composing it lie square to the forming dentine. In thin sections the odontoblasts form a single layer only, and in young teeth there is a distinct space between them. (Plate 36, fig. 2.) Dr. L. A. Weil ('Zur Histologie der Zahnpulpa') describes as a separate layer, a comparatively clear space existing between the odontoblasts and the general mass of the pulp tissue (Plate 36, fig. 1, d), in which there are no cells, but interlacing fine fibres, which he describes as being in connection with the odontoblast cells. This layer takes the stain very faintly, or not at all, generally appearing with low powers as a clear zone immediately beneath the odontoblasts.

I can certainly corroborate the fact of the constancy of this appearance in the transverse sections of teeth I have examined, although not agreeing with Dr. Weil as to the ultimate destination of all these fibres. In some longitudinal sections, however, taken near to the growing and unfinished end of a bicuspid tooth, I have been unable to distinguish any such layer, the cells and fibres of the general pulp tissue being in immediate proximity to the odontoblasts. In transverse sections near the apex of the pulp (where the growth of the dentine is not very active) I have seen the layer much more marked than in other parts. Dr. Weil

compares this layer to basal membranes (loc. cit.). Beneath this is seen the "gelatinous connective tissue" of the main substance of the pulp with its numerous cells, blood-vessels, and nerves (fig. 1, d).

In some of the first specimens which I cut by the balsam process (longitudinal sections of young healthy bicuspid teeth) my attention was arrested by a peculiar appearance at the border of the dentine which I had never seen in decalcified preparations.

Processes were seen springing from the dentine, and blending with the connective tissue of the pulp, all round the margin of the pulp cavity (Plate 36, figs. 3, 4, 5, 6). On examination with a higher power, these processes have the appearance of connective tissue bundles partially impregnated with lime salts in advance of the main line of calcification.

At the inner margin of the dentine they are seen to spring from its substance in a direction more or less parallel to the surface, these horizontal bundles of fibres blending together into larger bundles at right angles to the surface of the dentine, much as the spreading roots of a tree coalesce to form its trunk.

These bundles, the high refractive index of which suggests their partial calcification, are plainly seen to be continuous with the general connective tissue of the pulp. In the specimen from which the photographs* were taken, no stain was made use of, and in rubbing down the section much of the pulp was broken away from the dentine, rendering very conspicuous the connection of these fibres with the calcified tissue. I have not seen many specimens in which these connective tissue fibres gave evidence of partial calcification, and I am inclined to think the condition is an unusual one, although I have constantly, as described below, met with these fibres from the pulp incorporated with the portion of the matrix still unimpregnated with lime salts. (One is reminded, by these specimens, of the similar appearances in the formation of bone in membrane, where spiculæ are seen shooting out in advance of the calcified substance.)

The peculiar appearances exhibited by these specimens led me to examine other teeth for the same tissue, with the result that I found it was distinctly visible in the great majority of sections cut, the specimens above described being peculiar only in the large size and great apparent rigidity of these fibrous prolongations.

At the apex of the pulp cavity these processes are more slender, form wide, open loops, and can be traced for some distance into the pulp (Plate 36, fig. 6).

In sections cut somewhat obliquely (not in the same plane as the odontoblasts) the appearance shown in Plate 37, fig. 1, is often seen. Here small, deeply-stained cells, or cell-nuclei are seen, crowded upon, and following the course of, the bundles of connective tissue fibres, which in this specimen are very delicate. In other preparations, however, these bundles are much coarser, and the cells seem to be incor-

^{*} A set of photo-micrographs, illustrating this paper, are in the possession of the Royal Society. See p. 543 infra.

porated in the bundles (Plate 37, fig. 2), reminding one very much of the appearances in developing membrane-bone, where osteoblasts are seen applied to, and lying between, the bundles of osteogenic fibres.

In the last preparation referred to (Plate 37, fig. 2), which is cut in the plane of the odontoblasts, their nuclei are seen in the interspaces of these bundles although the outline of the cells cannot be made out.

A section which is not cut in the exact plane of the odontoblast cells exhibits the fibrous trabeculæ with greater distinctness than one cut in that plane, although they can readily be seen among the odontoblasts in the latter preparations. I think this is accounted for by the fact that the stained odontoblasts lying among these fibres hide them to a great extent, but when cut obliquely, the odontoblast nuclei are seen end on, and the regular layer of cells not being conspicuous, the connective tissue bundles are more clearly seen.

Many of the cells which are in contact with the processes above described, especially those which appear to be involved in the bundles, are distinctly smaller than the odontoblasts, and seem too closely applied to these bundles to be interpreted as odontoblasts. Associated, therefore, with these latter cells are other cells which, I believe, play an important part in dentine development, but they are destitute of processes, and not arranged in a definite layer.

In longitudinal sections of teeth where the tubes are cut obliquely, or nearly at right angles, especially near the upper part of the pulp cavity, the reticular structure exhibited in Plate 38, fig. 1, is sometimes visible, a fine net-work of fibres forming circular and oval meshes, involved in which are numerous small round cells. This appears to be the same reticulum of fibres described in the previous specimens, but seen, as it were, from beneath, the meshes being cut transversely.

Having constantly found these appearances in fully-formed teeth, I proceeded to examine specimens which had uncompleted fangs, in which, towards the unformed apex, the deposition of dentine was in active progress.

A longitudinal section from such a tooth exhibits the appearances shown in Plate 37, fig. 3.

In this rapidly growing portion of a tooth (Plate 36, fig. 2), the odontoblast cells are seen to be disposed in a single layer, to lie square against the layer of stained, uncalcified matrix substance (Plate 37, b), which is here very broad, and to be distinctly separated from one another. This slight separation between the odontoblasts I have found to be very constant in young dentine. A distinct reticulum of fine fibres is seen passing between and enveloping the odontoblasts, and by careful focusing on the right plane (see Plate 38, fig. 3, e), these fibres can be seen to be gathered into bundles and incorporated with the matrix substance out of which they appear to spring.

Small elongated and irregular shaped cells are seen in this specimen, mingled with the odontoblasts. In this section also, as in others of very young dentine which has not been decalcified, a faint striation parallel to the surface of the pulp cavity is visible in the recently calcified dentine.

This striation cannot be seen in these specimens in the layer between the odontoblasts and the dentine, but in the same layer in the incisor of a Calf kept for a long time in chromic acid, it can be seen very distinctly.

The striation in the dentine is limited to a narrow area, the matrix at some little distance from the pulp cavity showing no indication of it, neither is it visible in the last-formed layers of dentine near the apex of the pulp cavity at this stage of development. It is particularly evident in the dentine of the Rat.

Having satisfied myself of the presence of the connective tissue processes above described, in human dentine, I examined teeth of persistent growth, taking the incisor of the Rat (*Mus decumanus*).

A thin section, cut by Weil's process, shows a very strong connective tissue in the pulp, and a very open meshed reticulum of connective tissue bundles at the margin of the dentine, covered with small rounded cells similar in appearance to those of the main substance of the pulp. (Plate 37, fig. 4.)

In these specimens the incorporation of the connective tissue bundles with the forming dentine is particularly evident; the fibres, which form round and ovoid meshes at their point of junction with the dentine, lie in a horizontal or oblique position to the surface of the pulp cavity, and in many parts can be traced for a little distance into the substance of the formed dentine. Many of the small rounded cells seem to be incorporated with the fibres, as described above in human dentine, others are seen lying in the areolar spaces of the tissue.

The strongest fibrous bands are usually seen at a little distance from the growing base of the tooth, and the whole of this tissue towards the apex of the tooth was apparently considered by Mr. Tomes to be a degenerative tissue, as he says in the 'Dental Anatomy,' p. 367, "near to the surface actually in wear, they (the fibrils) become cut off from the pulp cavity by the conversion of what remains of the pulp into a laminated granular mass, so that the dentine exposed on the surface of a rodent's tooth must be devoid of sensitiveness." I find, however, that in sections cut by the balsam process the pulp contains odontoblasts as far forward as it extends, and that in all the specimens I have examined the tubules of the dentine pass through this laminated layer to the pulp tissue, and are nowhere cut off from connection with it and its cells. One would imagine that in the process of preparing such teeth by decalcification, the boundary between the laminated dentine and the laminæ of the pulp tissue would be obscured, and thus lead to such an interpretation as the above.

A very distinct striation of the dentine is noticeable at the margin of the pulp cavity, the individual striæ interlacing, but maintaining a general direction parallel to the connective tissue bands of the pulp which are incorporated with the matrix. These striæ are very visible for some way into the dentine, forming in this situation a

band of a slightly darker appearance than the rest of the dentine, and fading gradually away in the deeper parts of the tissue approaching the enamel. With good illumination the striæ can be detected in many parts in the form of fine lines in the deeper portion of the dentine. These markings bear a strong resemblance to those in a similar situation described above as sometimes seen in human teeth.

Allowing for the possibility of these connective tissue fibres being a degenerative tissue, I examined the growing base of the incisor of a young Rat, and found the same arrangement of fibres very distinctly blending with the dentine substance, and in this situation crowded with small polygonal cells, which in some parts were in such abundance that the septa seemed to be made up of cells (Plate 37, fig. 5).

The odontoblasts and the fibrous bands can in many parts be seen at the same time, but as in the case of the same tissue in human teeth, the fibres are most distinctly seen where the section is not cut exactly across the plane of the odontoblast cells.

In a molar tooth from a newly-born Rat, in which there was but a narrow strip of dentine formed, fine connective tissue fibres could be well seen running through the odontoblast layer to the dentine, and in some places forming loops at their junction with the dentine. Upon and around these are small cells, considerably smaller than the nuclei of the odontoblasts which are visible among them.

In some sections from the tusk of the Elephant, kindly forwarded to me by Professor MILLER of Berlin, I find a similar incorporation of the pulp tissue in the newly formed (tubular) dentine (Plate 38, fig. 2).

These specimens, however, having been prepared from dried pulps, do not exhibit the cells in their natural condition, although the passage of the connective tissue fibres of the pulp into the dentine is exceedingly well marked.

In the erratic deposits of secondary, non-tubular dentine, in the substance of the pulp of the tusk, so frequently met with, the incorporation of the stroma of the pulp in the tissue is very clearly seen (Plate 38, fig. 3).

At the suggestion of my friend, Mr. Charles Tomes, I examined the teeth of several fish in which an appearance of lamination is noticeable parallel to the surface of the pulp cavity, to discover if this same layer of connective tissue can be seen in vaso-dentine, between the formed dentine and the pulp. In the very characteristic vaso-dentine of the Hake (Merlucius), after many failures in consequence of the specimens not having been treated when sufficiently fresh, I obtained sections of these teeth showing a well defined layer around the pulp cavity, consisting of connective tissue fibres blending with the dentine, openings being present at intervals in this layer for the passage of the numerous blood-vessels (Plate 38, figs. 4 and 5).

In longitudinal sections these fibres are seen lining the pulp cavity from base to apex of the tooth; they are seen to be in close apposition to the most recently formed layer of vaso-dentine matrix, and have a very well defined limit towards the pulp.

The individual fibres when examined with a high power, are wavy in outline, many of them being somewhat flattened; they show a tendency to form arches with one

another, and the interval between the concavities of these fibres is often occupied by two or three curved ones passing from one side to the other. Their termination towards the pulp appears abrupt, but with high powers they are seen to be attached to the stroma of the pulp by delicate fibres. This layer is very closely applied to the blood-vessels, and in many places where a horizontal vessel traverses the pulp close to the dentine, these fibres appear to be attached to the vessel, or rather, it is involved in the meshes of the delicate pulp tissue passing from this layer to the deeper portion of the pulp. Here and there, one of the large flattened fibres may be seen to extend beyond the others, deeply into the pulp, there dividing into several branches. An unstained specimen shows very faint traces of cells near the dentine, but in those stained with carmine a layer of polygonal cells is visible in close apposition to the These cells vary much in size; they have a coarsely granular appearance, with a well-marked nucleus, and take the stain deeply. Near the base of the tooth they form a well-marked layer and are placed closely together (Plate 39, figs. 1 and 2), but as the apex is approached they have a more rounded outline and less definite arrangement. Many of these cells have short processes (Plate 39, fig. 3) passing from one cell to the other, and here and there to the dentine, and very similar cells are seen scattered throughout the pulp.

From their close application to the dentine, their regular arrangement, and their strong resemblance to osteoblasts, I believe these cells to be intimately concerned in the calcification of the vaso dentine matrix. At the base of the anchylosed tooth of the Hake, cells precisely similar in appearance are seen in contact with the surface of the bone.

In an important paper by Mr. Charles Tomes "On the Structure and Development of Vascular Dentine," published in the 'Philosophical Transactions of the Royal Society' for 1878 (vol. 169, Part I.), he gives an account of the mode of development of this tissue which has since been generally adopted. The author describes a layer of odontoblasts as "clothing the whole pulp, and where there is a capillary at the surface they clothe *it*, so that when they calcify, the capillary becomes solidly imbedded in the dentine."

From an examination of the specimens which I have prepared, as described above, I am inclined to think that much of the tissue here called odontoblasts consists of the connective tissue I have alluded to.

Mr. Tomes has kindly lent me some of his specimens, preserved in balsam and in glycerine jelly. In these latter preparations the great majority of the processes attached to the blood-vessels are precisely similar in appearance to the fibres I have described. Mr. Tomes notices in his paper that the oval nucleus of the odontoblast cells is in some cases very distinct, in others indistinguishable, and says: "I have been unable to discover in what conditions this is the case." The explanation I believe is in the fact that in the teased-out specimens, the cells which play the part of odontoblasts lie here and there in the meshes of this connective tissue. Many of

the flattened fibres look very like cells, but are distinguished by the absence of a nucleus.

I think it is these fibres which clothe the blood-vessels that are in close apposition to them in their course through the pulp tissue, and it appears highly probable that in teasing out the pulp in any fluid medium, the blood vessels would be likely to draw away with them this layer of fibres from its attachment to the dentine.

In some transverse sections which were prepared by Mr. Tomes and stained with hæmatoxylin, the layer of connective tissue fibres is deeply coloured, and where these are crowded together they look very like odontoblasts, but thin sections show them to consist of fine fibres and also show the absence of nuclei at their distal extremities.

While therefore agreeing with him as to the existence of an odontoblast layer in vaso-dentine, I think that the cells are very different in form to the odontoblasts of Mammalian teeth, bearing much more resemblance to osteoblasts, and that they lie in the meshes of, and are surrounded by a layer of, connective tissue fibres in intimate connection with the dentine.

These fibres form a much more definite and sharply defined layer than in any Mammalian teeth, so much so that they have been regarded by the above-mentioned author as a peculiar odontoblast layer.

After a reconsideration of his own and of my specimens, Mr. Tomes agrees with me that they must be considered to be of the nature of a connective tissue.

Some of his more recent investigations on the dentine of the Cod have a strong bearing on this subject. He finds an outer marginal layer of dentine in which bundles, like connective tissue bundles in a hyaline matrix, are plainly seen, springing from the deeper vascular part of the dentine; these fibres are arranged radially like those surrounding the pulp cavity. Also in transverse sections of the decalcified Hake's tooth the dentine is often seen split up into fine fibres, likewise arranged at right angles to the surface of the pulp cavity (Plate 39, figs. 4 and 5).

There still remains the not inconsiderable difficulty that the layer of connective tissue which I have described as surrounding the pulp of these vaso-dentine teeth, which is sharply defined and of great regularity, consists only of radial fibres; so that we are still no nearer to an explanation of the *concentric* striation of vaso-dentine.

As a result of the above observations I proceeded to examine teeth for evidences of lamination in the completed dentine, or for any indications of its having been developed on a connective tissue basis.

In corroboration of this latter point, connective tissue fibres are sometimes brought into view in the substance of the dentine in teeth softened by caries, the acid formed in the progress of this disease dissolving out the lime-salts.

Close to the inner margin of the dentine in a case in which caries had encroached upon the pulp cavity, the appearance represented in Plate 36, fig. 7, is visible. Here

the lime-salts and the tubules seem to have been dissected away, as it were, by the acid, exposing the connective tissue basis of the matrix.

As evidence of the completed dentine retaining something of a laminar structure, the following arguments may be adduced:—

- 1. Teeth decalcified by the mineral and other acids, break up at right angles to the tubes, that is concentrically with the pulp cavity.
- 2. Teeth partially decalcified by treatment for a short time with acids sometimes exhibit this splitting in a very marked degree.
- 3. There is a faint appearance of striation (before referred to) in many teeth cut by the balsam process, this appearance being confined to the portion of the dentine which has been most newly formed, nearest to the active odontoblast cells in rapidly growing teeth.
- 4. Mr. F. J. Bennett, in a paper read before the Odontological Society in 1888,* described the appearances produced by the action of glycerine on dentine in which laminæ were brought into view.
- 5. In several specimens of carious teeth, in the portion of dentine yet uninvaded by micro-organisms, but within the area of partial decalcification in advance of them, a minute striation is visible, bearing a very strong resemblance to the striation of voluntary muscular fibres, interrupted here and there by some rounded contours which it is somewhat difficult to explain.
- 6. In the vaso-dentine of fish, besides the lamination parallel to the surface of the pulp cavity, the dentine in decalcified specimens splits up at right angles to the pulp cavity, and bundles of fibres are seen following the same direction incorporated with the outer layers of dentine in the Cod (above referred to).

Now, unless it be held that these appearances have been brought into view by the processes employed, which I think can hardly be maintained by anyone who examines the specimens, the verification of these observations must lead to some modification of the ordinarily received views of dentine development.

We can no longer look upon the matrix of dentine as being a homogeneous substance, but must regard it as composed of a reticulum of fine fibres of connective tissue modified by calcification, and where that process is complete, entirely hidden by the densely deposited lime salts. These fibres decussate freely with one another, and I believe them to be analogous to the decussating fibres of bone. They are rendered visible in some instances by the slow decalcifying action of caries, as they appear to resist the action of acids more than do the lime salts.

For this layer of connective tissue surrounding the pulp and entering into the substance of the matrix I would suggest the term "odontogenic fibres," from their great similarity to the osteogenic fibres of bone. There are objections to the term osteogenic, as applied to these fibres in bone, as they are the scaffolding on which the bone is built up, rather than actually the genetic tissue of the bone, and the same objection

^{* &#}x27;Transactions of the Odontological Society of Great Britain,' vol. 21, November.

holds good for the term odontogenic, but it is suggested as the most convenient appellation, and the one by which their analogy to the similar fibres in bone is best indicated.

In the formation of sub-periosteal bone, bundles of fibres from the periosteum are seen springing from the bone, and in the alveoli formed between these bundles the osteoblasts lie.

These periosteal fibres penetrate the bone, and can be distinctly seen in it in many places, although the great majority of them are afterwards obliterated by absorption of the bone first formed, and the formation of dense bone around the Haversian canals—the so-called Haversian systems.

A similar appearance is seen in cementum, the fibres being visible in this tissue as Sharpey's fibres. (Plate 37, fig. 6.)

The above investigations as to the occurrence of this tissue surrounding the pulp cavity in teeth, suggest the view that these fibres are the scaffolding on which the tooth matrix is built up—just as calcification proceeds in bone along and around the osteogenic fibres—that they are incorporated in the matrix of the dentine by calcification, and form really the basis of its substance.

In the 'Dental Anatomy' (p. 62), Mr. Tomes says, "Several varieties of dentine exist in which those peculiarities of structure which differentiate it from bone are less marked, so that a point is sometimes reached at which it is hard to say whether a particular structure should more rightly be regarded as dentine or as bone. Hard or tubular dentine has always been considered least like bone in structure and in development."

Again, on p. 176 (loc. cit.), speaking of the development of osteo-dentine, the same authority says, "With the exception of the thin external layers, which are developed from a superficial layer of not very highly specialised cells, osteo-dentine is built up in a manner fundamentally different from that in which hard dentine, plici-dentine and vaso-dentine are constructed. Its inner surface becomes roughened by trabeculæ shooting inwards into the substance of the pulp, which speedily becomes traversed completely by them, as well as by the connective tissue bundles which are continuous with them. Osteoblasts clothe, like an epithelium, the trabeculæ and the connective tissue fibres attached to them, and by the calcification of these the osteo-dentine is formed. The process is exactly like calcification of any membrane bone, and the connective tissue bundles remind one of those which are believed to be the occasion of the formation of Sharpey's fibres in bone."

The appearances in dentine which I have described in the earlier part of this paper, would seem to point to a mode of development of hard or tubular dentine which in many essential points tallies with the description of the development of osteo-dentine above given, and consequently presents a strong analogy to the development of bone in membrane.

In human dentine, as I have shown, trabeculæ are seen shooting inwards into the

pulp from the surface of the forming dentine. These trabeculæ (sometimes exhibiting an appearance as if stiffened by the deposit of lime-salts in advance of the general line of calcification) are continuous with the connective tissue fibres of the pulp.

These fibres and trabeculæ are also in the case of hard dentine, as in that of osteodentine and bone, covered with cells, which in many parts thickly clothe them, and which, it is to be supposed, have similar functions to osteoblasts. Smaller cells are intimately associated with the odontoblasts proper, the latter cells being also involved in the connective tissue stroma in continuity with the dentine, and according to the view which under the circumstances seems most reasonable, these cells together secrete a material which calcifies along the lines of the odontogenic fibres.

This view appears to be supported by the investigations into the development of vaso-dentine above recorded, for here the cells at the margin of the pulp cavity bear a strong resemblance to osteoblasts, and lie between and among the connective tissue fibres springing from the matrix, just as the osteoblasts lie between the connective tissue bundles in the calcification of sub-periosteal bone.

While convinced that the views I have advanced are correct as far as they go, I acknowledge that they render some things perhaps more difficult of explanation than do those ordinarily received; for example, the nature of the dentinal fibrils, their relation to the odontoblasts, and the share taken by them in dentine formation, were more easily explained upon the hypothesis that the matrix was formed by the direct conversion of a portion of the odontoblast. Further investigation into the contents of the dentinal tubes seems called for; indeed, the usually accepted view of the origin of the fibril has not appeared satisfactory to all observers. Professor Klein says, "However great the authorities who maintain that the cells of the outer stratum above referred to as the odontoblasts proper, send processes into the dentinal canals as the dentinal fibres, I must question the accuracy of this assertion, for I cannot find convincing evidence of those odontoblasts doing more than producing the dentine matrix the dentinal fibres appear to me to be derived solely from the deeper layer of cells which are wedged in between the former."—'Atlas of Histology.'

The same view was maintained by Dr. Andrews, of Boston, in a paper read before the IXth International Medical Congress at Washington.

My own observations on the relations of the dentinal fibril are as yet incomplete, many appearances met with being somewhat contradictory.

As pointed out by many observers, there is always present in developing dentine a layer of tissue between the odontoblasts and the fully calcified matrix which is "on the borderland of calcification," a tissue believed by those who hold the conversion view, to consist of the consolidated masses of odontoblasts prior to their calcification, but which according to the view of secretion here maintained, is a material elaborated by the odontoblasts and other cells upon a connective tissue foundation. It appears probable that this tissue being gradually saturated by the lime-salts elaborated by the cells, becomes supersaturated at a certain distance from the secreting cell, a

process analogous to crystallisation* takes place, and the globules of calcoglobulin are deposited.

The calcification of the matrix of tubular dentine is so complete that the fully formed tissue appears to be perfectly homogeneous.

A faint striation in newly formed dentine may, however, be detected, and is very marked in the incisor of the Rat before referred to; moreover, the effects of artificial decalcification and that caused by the progress of caries are such as to render visible this indication of lamination, and point to the original development of the matrix on a connective tissue foundation.

To Mr. Charles Tomes I am much indebted for the kind assistance and valuable suggestions he has given me throughout this investigation, and my thanks are also due to the Directors of the Marine Biological Association for the kind manner in which they have supplied me with fresh specimens, preserved by special methods.

Note.

Added March 5th, 1891.

Since communicating this paper, I have received from Vienna a paper by Professor V. von Ebner, which has a considerable bearing on the subject of the present communication. It is entitled "Histologie der Zähne mit Einschluss der Histogenese," and has been published recently in the 'Handbuch der Zahnheilkunde' now appearing in separate parts (Vienna, 1890–91).

Professor von Ebner says: "The dentine matrix appears to be homogeneous in sections ground in water, whilst in bone a fibrillar structure may be observed. If, however, dentine is decalcified with hydrochloric acid in a 10 or 20 per cent. solution of common salt, one can detect in fine sections, or in thin pieces scraped off with a knife, a fibrillar structure. In pieces that have been torn off, we sometimes obtain the fibrillæ isolated. These fibres are exceedingly fine, scarcely more than 5 μ thick" (the diameter of the dentinal fibril being from 1.3 μ to 2.5 μ). "On the whole they are very similar to those of bone and also to those of fibrous tendinous tissues.

"They swell up in alkalies and acids; they are uniaxial and double-refractive; in short, in everything they present the same characteristics as the glue-giving connective tissue fibres. The fibres, as in typical bone, are united into bundles about 2μ in diameter, not however arranged in lamellæ as in bone. The principal direc-

* Von Ebner considers the optical characters of *Enamel* and its behaviour under the action of weak acids as strong evidence of its crystalline character. Von Ebner, 'Sitzungsberichte d. Kaiser. Akad. d. Wissenschaften, Wien,' 1889.

tion of the bundles corresponds to the longer axis of the tooth, being by no means, however, parallel to it. On the other hand the bundles cross each other, and mostly in planes perpendicular to the dentinal tubules."

This author also refers to a laminar structure occasionally exhibited here and there in carious dentine, but he does not enter on the subject of dentine development or refer to any connection between the dentine and the connective tissue of the pulp; the appearances he describes, however, in formed dentine after decalcification will, if confirmed by other observers, go far to establish the truth of the views suggested in the present paper.

DESCRIPTION OF PLATES 36-39.

PLATE 36.

- Fig. 1. Human bicuspid tooth at age of 14. Transverse section of tooth and pulp prepared by Well's method. (a) Dentine; (b) layer of tissue "on the borderland of calcification"; (c) odontoblast layer (the nuclei of several layers of cells visible); (w) the clear zone, with fine fibres, described by Dr. Well; (d) pulp, with cross sections of several blood-vessels. Magnified 75.
- Fig. 2. Human bicuspid tooth, which had been only partially erupted, the apex of the root not being completed. (a) The last calcified portion of the dentine; (b) the layer described by the same letter in fig. 1; this layer is much wider in proportion to the dentine than in teeth in later stages of development. Magnified 250.
- Figs. 3, 4, and 5. Fibres springing from the dentine along the pulp margin of a Human bicuspid tooth, longitudinal section. These fibres appear to be stiffened by calcification in advance of the mass of the dentine. Magnified 350.
- Fig. 6. Fibres of the connective tissue of the pulp in continuity with the layer of the dentine which has not received its full deposit of lime salts; at the apex of the pulp cavity of a bicuspid tooth (Human) longitudinal section.

 Magnified 350.
- Fig. 7. Human molar tooth softened by caries, showing fibres in the substance of the dentine. Magnified 200.

PLATE 37.

Fig. 1. Fine fibres in connection with the dentine on one side and the pulp on the other, which are here seen to be crowded with cells (or cell nuclei).

Transverse section of pulp of crown of a bicuspid tooth (Human). Magnified 230.

- Fig. 2. Human bicuspid. Transverse section of the pulp of the crown. Processes covered with cells. Larger cells lying in the spaces between these fibres, which appear to be the nuclei of odontoblasts. Magnified 230.
- Fig. 3. Human bicuspid tooth with uncompleted root. Longitudinal section a short distance from the open end of the root. (d) Dentine exhibiting indications of transverse striation, with the advancing line of rounded and globular masses of calcoglobulin; (b) the layer on the border land of calcification; (e) fine fibres of the pulp blending with the last layer of the dentine (b); (o) the odontoblast layer, not fully in focus; (p) the pulp, with its fusiform and other cells, some of which are seen lying among the odontoblasts. Magnified 230.
- Fig. 4. From a longitudinal section of tooth (incisor) of Rat (Mus decumanus). A narrow portion of newly-formed dentine at the growing base of the tooth.

 (a) Dentine;
 (b) uncompleted layer of dentine;
 (f) connective tissue incorporated with dentine, and in this position crowded with small cells. Magnified 230.
- Fig. 5. Incisor of Rat (Mus decumanus), longitudinal section at about the middle of the length of the tooth; (d) dentine showing very marked transverse striation, the general direction of these striæ being slightly oblique, in the same direction as the connective tissue fibres of the pulp (f) incorporated with the dentine. Magnified 175.
- Fig. 6. Cementum from transverse section of Human bicuspid tooth. (g) Granular layer; (s) fibres of Sharpey penetrating the outer layer of the cementum; (p) peridental membrane. Reduced from Camera lucida drawing, $\frac{1}{16}$ th oil immersion (Powell and Leland). Magnified 500.

PLATE 38.

- Fig. 1. Human molar tooth, oblique section. Appearance of a reticulum of fibres at the pulp margin of the dentine forming very rounded meshes and studded with small round cells. Magnified 350.
- Fig. 2. From transverse section of tusk of Elephant. (a) Ivory; (c) connective tissue of the pulp, prepared from a dry pulp (Dr. MILLER's specimen). Magnified 350.
- Fig. 3. Secondary deposit in pulp of Elephant's tusk. (α) The calcified nodule;
 (c) connective tissue of pulp. Magnified 350.
- Fig. 4. Longitudinal section of tooth of Hake (Merlucius vulgaris), showing at (d) vaso-dentine traversed by (b) blood vessels; (f) layer of connective tissue fibres surrounding the pulp. Magnified 150.
- Fig. 5. Portion of the same more highly magnified (700 diameters), to show the connective tissue fibres (f), and their relations to the dentine (d), and to the pulp and blood-vessels (b).

PLATE 39.

- Fig. 1. Vaso-dentine (*Merlucius vulgaris*); (d) vaso-dentine traversed by blood-vessels; (c) a layer of cells in close apposition to the dentine. Magnified 350.
- Fig. 2. The same cells more highly magnified. Magnified 700.
- Fig. 3. Some of the same cells at the base of the tooth, exhibiting processes.

 Magnified 700.
- Fig. 4. From a transverse section of tooth of Hake, decalcified (Mr. Tomes' specimen); (d) vaso-dentine splitting into fibres; (c) layer of connective tissue fibres in pulp; (b) a blood-vessel. Magnified 75.
- Fig. 5. From the same preparation as fig. 4, more highly magnified. Magnified 700.

DESCRIPTION OF UNBUBLISHED PHOTO-MICROGRAPHS DEPOSITED WITH THE ROYAL SOCIETY.

No. I.

- Fig. 1. From a transverse section of a bicuspid tooth (Human), prepared by Weil's balsam process, stained with borax-carmine. (a) Dentine; (b) tissue "on the border-land of calcification"; (c) pulp with odontoblast layer. Magnified 200 diameters, \(\frac{1}{4}\)-inch objective (SWIFT).
- Fig. 2. Transverse section of one cornu of the pulp of a young bicuspid tooth (Human). Magnified 75 diameters, ½-inch objective; stained with borax carmine.

No. II.

- Process or bundles of fibres which are seen to spring from the dentine (a) and in the calcified portion of which they appear incorporated; from the margin of the pulp cavity of a Human bicuspid tooth (age 14), unstained.
- Fig. 1. Magnified 200 diameters, $\frac{1}{4}$ -inch objective (SWIFT).
- Fig. 2. One of the processes more highly magnified, 500 diameters, Powell and Leland oil immersion $\frac{1}{16}$.

No. III,

Other similar bundles from the same preparation. Magnified 320 diameters, Zeiss apochromatic objective 3 mm., dry.

No. IIIa.

- Fig. 1. Similar bundles at apex of pulp cavity. Magnified 320.
- Figs. 2 (magnified 200) and 3 (magnified 75). From the middle of the length of the tooth.

No. IV.

- Fig. 1. From the margin of the pulp cavity in the crown of a Human bicuspid (age 14).

 (a) Dentine; (b) fine fibres between the dentine and the main body of the pulp, crowded with nuclei near the dentine. Magnified 320 diameters, Zeiss apochromatic 3 mm.
- Fig 2. Reticular appearance at the margin of the dentine in a Human molar tooth (longitudinal section), the dentine is cut obliquely, and viewed from beneath; numerous cells are seen upon the septa and lying in the interspaces. Magnified 200 diameters, 4-inch objective (Swift). (This preparation is not a suitable one for photography and is better represented by the drawing, Plate 38, fig. 1).

No. V.

Human bicuspid tooth. Transverse section. Showing processes in the pulp (b) connected with the dentine (a) and crowded with cells.

Figs. 1 and 2. Magnified 200 diameters, \(\frac{1}{4}\) inch (SWIFT).

No. VI.

Figs. 1 and 2. Human tooth (bicuspid). Longitudinal section close to uncompleted apex of growing tooth. (a) Newly calcified dentine showing minute transverse markings, more plainly seen in fig. 3; (b) layer of uncalcified matrix (in this young condition of the tooth of great proportional width); (c) pulp with large odontoblast and other cells. (In the photographs the objective has been focussed on the connective processes at the margin of the dentine, the odontoblasts being slightly out of focus.) Zeiss apochromatic 3 mm. Magnified 320 diameters.

No. VII.

Figs. 1 and 2. Longitudinal section of incisor tooth of Rat (Mus decumanus),
(a) Enamel; (b) dentine; (c) lamination in dentine; (d) connective tissue of pulp incorporated in the dentine. Magnified 250 diameters, \(\frac{1}{4}\)-inch objective (SWIFT).

No. VIII.

Fig. 1. Transverse section from tusk of Elephant, from a dried specimen, afterwards softened and mounted in balsam (Dr. Miller's specimen). Magnified 320 diameters. Zeiss apochromatic 3 mm. objective.

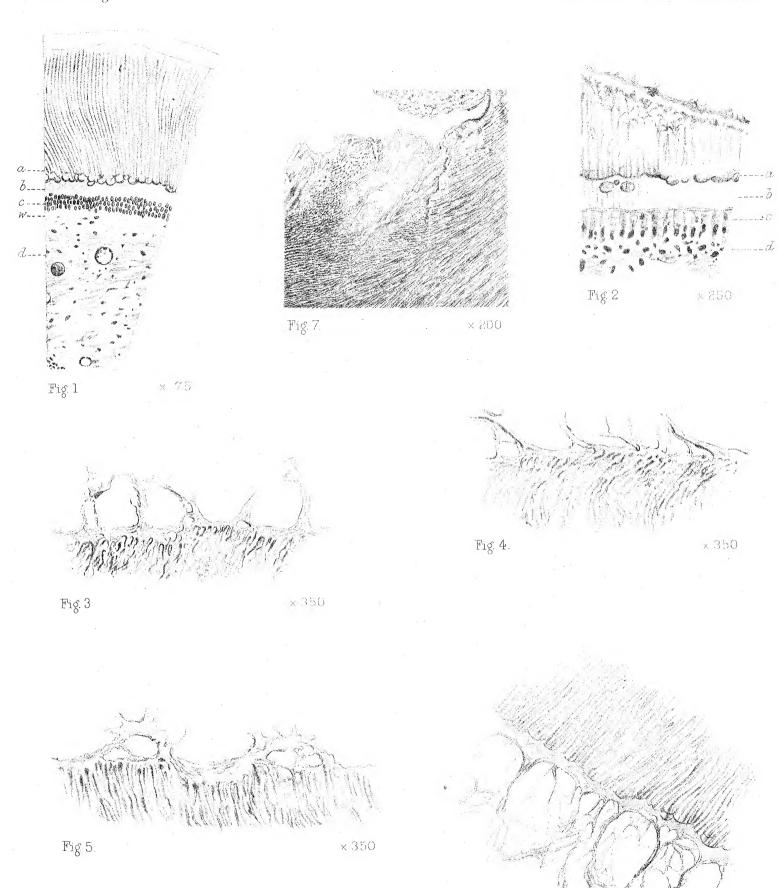
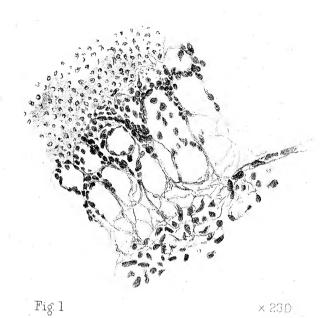
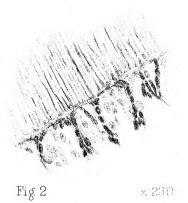
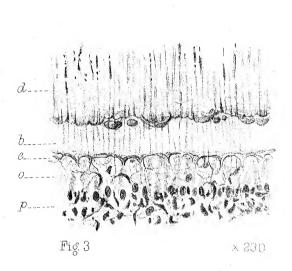


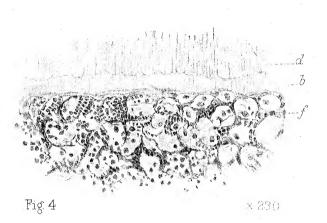
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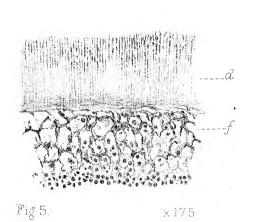
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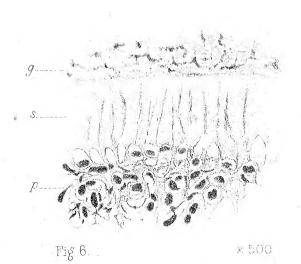


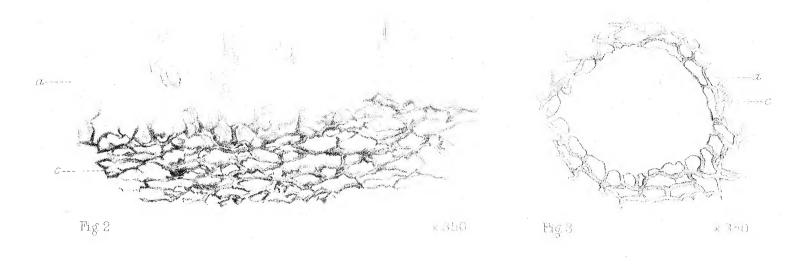












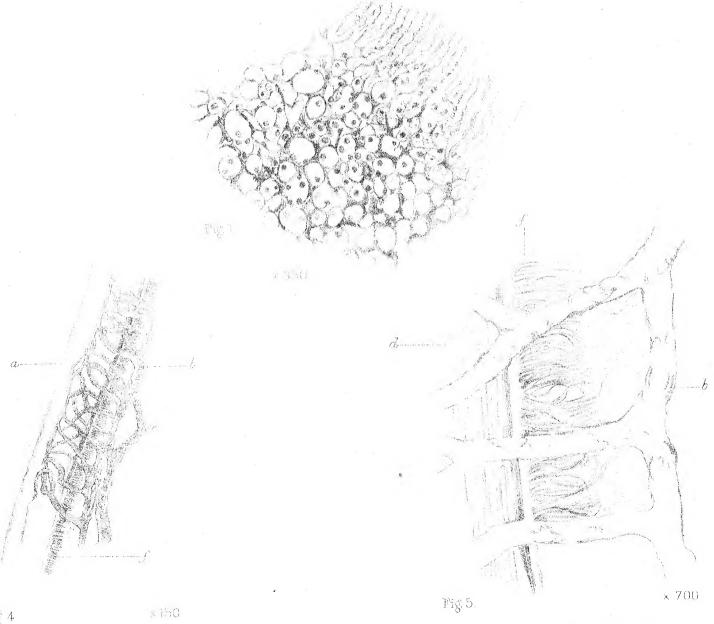
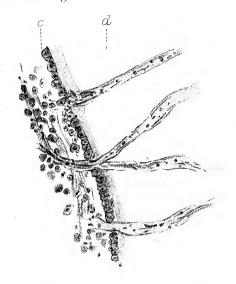


Fig 4





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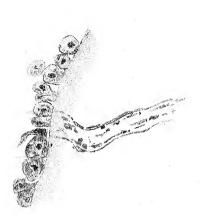


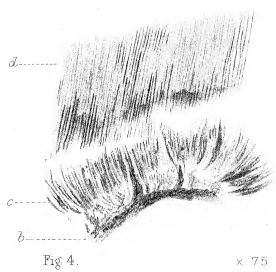
Fig. 2.

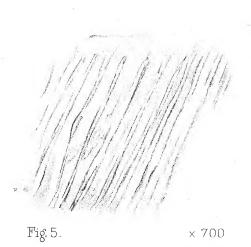
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Fig. 3.

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× 700

Fig. 2. Pulp-stone or secondary deposit from pulp of same tusk, showing the incorporation of the connective tissue of the pulp with this tissue. (d) Dentine; (p) pulp. Magnified 320 diameters, Zeiss apochromatic 3 mm. objective.

No. IX.

Vaso-dentine. Tooth of Hake (Merlucius vulgaris).

- Fig. 1. Longitudinal section. (a) Vascular dentine traversed by blood-vessels; (b) layer of connective tissue fibres in contact with the dentine and lining the pulp cavity. Magnified 300 diameters, 4 mm. apochromatic (Zeiss).
- Fig. 2. From the margin of the pulp cavity at the base of the tooth of a Hake—showing cells, many of which have short processes lying upon the pulp surface of the dentine. (α) Dentine with blood-vessels; (b) cells. Magnified 320 diameters, 3 mm. apochromatic (Zeiss).

No. X.

- Fig. 1. Appearance of fibres in the substance of the dentine near the pulp cavity in a molar tooth (Human) softened by caries. Magnified 200 diameters, \(\frac{1}{4}\)-inch objective (SWIFT).
- Fig. 2. Transverse splitting of human tooth at right angles to the tubes, caused by the action of formic acid. Magnified 175 diameters, $\frac{1}{4}$ inch.

No. XI.

- Fig. 1. Root of Human bicuspid, transverse section. (a) Granular layer of cementum; (b) stained layer showing Sharpey's fibres; (c) cells and tissue of peridental membrane. Magnified 500 diameters, Powell and Leland 16.
- Fig. 2. Transverse markings in the dentine in caries within the zone of decalcification. Magnified 500 diameters, Powell and Leland 16 oil immersion.

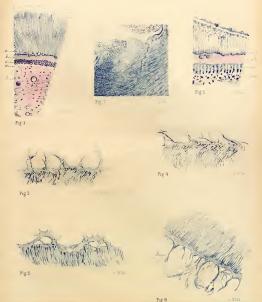


PLATE 36.

- Fig. I. Human bicupid tooth at age of 14. Transverse section of tooth and pulp prepared by Wint's method. (a) Dentine (b) byter of issues "on the borderland of eachigantion"; (c) obnotablast layer (the nuclei of several layers of cells visible); (e) the clear zone, with fine fibres, described by Dr. Wunt; (d) pulp, with cross sections of several blood-vessels. Magnified 75.
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- Figs. 3, 4, and 5. Fibres springing from the dentine along the pulp margin of a Human bicuspid tooth, longitudinal section. These fibres appear to be stiffened by calcification in advance of the mass of the dentine. Magnified 350.
- Fig. 6. Fibres of the connective tissue of the pulp in continuity with the layer of the dentine which has not received its full deposit of lime salts; at the apex of the pulp cavity of a bicuspid tooth (Human) longitudinal section. Magnified 350.
- Fig. 7. Human molar tooth softened by caries, showing fibres in the substance of the dentine. Magnified 200.







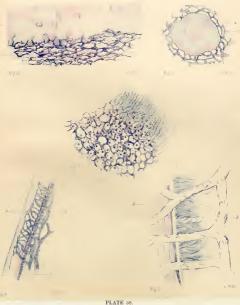






PLATE 37.

- Fig. 1. Fine fibres in connection with the dentine on one side and the pulp on the other, which are here seen to be crowded with cells (or cell nuclei). Transverse section of pulp of crown of a bicuspid tooth (Human). Magnified
- Fig. 2. Human bicuspid. Transverse section of the pulp of the crown. Processes covered with cells. Larger cells lying in the spaces between these fibres, which appear to be the nuclei of odontoblasts. Magnified 230.
 Fig. 3. Human bicuspid tooth with uncompleted root. Longitudinal section a short
- distance from the open end of the root. (d) Destine exhibiting indications of transverse stration, with the advancing line of rounded and globular masses of caleoglobelin; (d) the layer on the border land of calcification; (e) fine fibres of the pulp blending with the last layer of the dentine (b); (i) the odoutoblast layer, not fully in foous; (p) the pulp, with its fusifiers and other cells, some of which are seen lying among the doutoblasts. Magnified 230.
- Fig. 4. From a longitudinal section of tooth (incisor) of Rut (Mas decomment). A narrow portion of newly-formed dentine at the growing base of the tooth. (a) Dentine; (b) uncompleted layer of dentine; (f) connective these incorporated with dentine, and in this position crowded with small cells. Magnified 230.
- Fig. 5. Incisor of Rat (Mus decumanus), longitudinal section at about the middle of the length of the tooth; (d) dentine showing very marked transverse striation, the general direction of these strine being slightly oblique, in the same direction as the connective tissue fibres of t'ie pulp (f) incorporated with the dentine. Magnified 175.
- Fig. 6. Cementum from transverse section of Human bicuspid tooth. (g) Granular layer; (s) fibres of Shardery penetrating the outer layer of the cementum; (p) peridental membrane. Reduced from Camera lucida drawing, 12th oil immersion (Powell and Leland). Magnified 566.



- Fig. 1. Human molar tooth, oblique section. Appearance of a reticulum of fibres at the pulp margin of the dentine forming very rounded meshes and studded with small round cells. Magnified 350.
- Fig. 2. From transverse section of tusk of Elephant. (a) Ivory; (c) connective tissue of the pulp, prepared from a dry pulp (Dr. Miller's specimen). Magnified 350.
- Fig. 3. Secondary deposit in pulp of Elephant's tusk. (a) The calcified nodule; (c) connective tissue of pulp. Magnified 350.
- Fig. 4. Longitudinal section of tooth of Hake (Merlucius vulgaris), showing at (d) vaso-dentine traversed by (b) blood vessels; (f) layer of connective tissue fibres surrounding the pulp. Magnified 150.
- Fig. 5. Portion of the same more highly magnified (700 diameters), to show the connective tissue fibres (f), and their relations to the dentine (d), and to the pulp and blood-vessels (b).











PLATE 39.

- Fig. 1. Vaso-dentine (Merlucius vulgaris); (d) vaso-dentine traversed by blood-vessels; (e) a layer of cells in close apposition to the dentine. Magnified 350.
- Fig. 2. The same cells more highly magnified. Magnified 700.
 Fig. 3. Some of the same cells at the base of the tooth, exhibiting processes.
- Magnified 700.

 Fig. 4. From a transverse section of tooth of Hake, decalcified (Mr. Tomrs' specimen);
 (d) vaso-dentine splitting into fibres; (e) layer of connective tissue fibres in
- pulp; (b) a blood-vessel. Magnified 75.
 Fig. 5. From the same preparation as fig. 4, more highly magnified. Magnified 700.